2 Fluid Physiology Part 1: Volume and Distribution of Water and Its Major Solutes Between Plasma, the Interstitium and Intracellular Fluid

Thomas Woodcock

Contents

T. Woodcock (✉)
Department of Critical Care, University Hospital Southampton, Southampton, UK

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IFA Commentary (MLNGM)

In this chapter, we undertake a deep dive into the secrets of the different fluid spaces and learn about body composition. How many body compartments exist? This question can be dealt with in different ways, classically there are fat tissue—water—proteins—and minerals while water on its own is also distributed into four compartments: intracellular water—interstitial water—intravascular water—and transcellular, with extracellular water calculated as the sum of interstitial plus intravascular plus transcellular water content (Fig. 2.1). There is ongoing discussion about the different fluid spaces and there are traditionally four: the first or intravascular space—the second or interstitial space—the third or pleural and peritoneal space—and the fourth space or the transcellular fluid. And not to forget the lymphatic system. Each compartment or space is separated from each other and the surrounding fluids by specific cells and membranes and an endothelial glycocalyx layer surrounding the vascular space, which regulates fluid and electrolyte shifts and transport between different compartments, spaces and cells. The endothelial glycocalyx is a thin negatively charged proteinaceous mesh-like layer, a gel-like matrix that surrounds all vascular endothelium on the luminal surface. It is composed of membrane-bound glycoproteins and proteoglycans. It was previously thought to be inert but plays a key role in vascular integrity and function: the regulation of vascular permeability, endothelial anticoagulation and modulation of interactions between the endothelium and the vascular environment. Thus, it prevents the free movement of water and electrolytes. Disruption or degradation of the glycocalyx may be an important mediator of inflammation, oedema, sepsis syndromes and capillary leak syndromes. Therefore, in various surgical and disease states, the glycocalyx has the potential as a novel therapeutic target. These new insights gave birth to an updated version of the traditional Starling equation that incorporates the current understanding of the role of the endothelial glycocalyx in transvascular fluid filtration, also known as the Starling Principle. The traditional principle describes the fluid passage across the semipermeable capillary membrane, which is determined by the net result between hydrostatic and oncotic pressures such that the fluid leaves the capillary at the

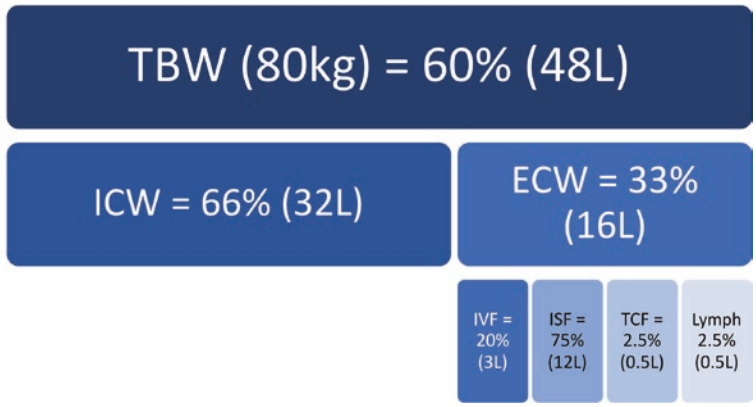

Fig. 2.1 Body water composition for an adult 80 kg male. *ECW* extracellular water, *ICW* intracellular water, *ISF* interstitial fluid, *IVF* intravascular fluid, *TBW* total body water, *TCF* transcellular fluid

arterial end of the capillary and is absorbed at the venous end of the capillary. The Revised or extended Starling Principle recognises that, because microvessels are permeable to macromolecules, a balance of pressures cannot halt fluid exchange. In most tissues, steady oncotic pressure differences between plasma and interstitial fluid depend on low levels of steady filtration from plasma to tissues for which the Revised Principle provides the theory [1].

Learning Objectives

After reading this chapter, you will:

1. Learn about interstitial fluid dynamics in order to manage clinical problems of fluid and albumin maldistribution.
2. Understand that lymphangions propel lymph flow, an active and vital part of the circulation of extracellular fluid and soluble proteins including albumin from capillary beds to the great veins.
3. Learn that the lymphatic and immune cell systems have an important role in the storage and release of sodium ions.
4. Understand that the relationship between intravenous sodium dose and oedema is not as clear cut as previously believed.
5. Comprehend that the separation of cytosol from extracellular fluid includes volume-regulated anion channels within cell membranes, enabling intracellular fluid volume to be maintained in the face of wide variations in extracellular fluid composition.
6. Know that the prescription of adequate amounts of potassium to patients is important.

Introduction

An appreciation of the distribution of water (the aqueous biological solvent) and its solutes is fundamental to understanding the physiology of body fluid spaces. The normal compartmentalisation of body water has been measured in various ways, and the decompartmentalisation seen in many critical illnesses is held to be an important pathophysiological phenomenon. Traditionally blood has been treated as an intravascular fluid circulation, while the lymphatic vessels have been called a drainage system. The modern view is of an actively pumped double circulation of extracellular fluid (intravascular plasma, and interstitial fluid and lymph) that enables vital solutes to be transferred to and from the intracellular fluid. Fluid flux across cell membrane barriers and across the different microvascular permeability barriers is determined by hydrostatic pressure differences and solute concentration gradients. Total body water volume is largely regulated by the pituitary hormonal effect of arginine vasopressin acting on vasopressin type 2 receptors in renal collecting ducts to retain or release water. In critical illness, arginine vasopressin deficiency predisposes patients to dilutional hyponatremia if the infused volume is larger than necessary or excessively prescribed as 'electrolyte-free water'. Body sodium is conserved by the renin–angiotensin–aldosterone axis regulating the degree of sodium reabsorption in renal distal tubules. Body sodium largely determines the proportion of body water that comprises extracellular fluid volume, but there is significant non-osmotic sodium storage capacity in the interstitium, particularly in the interstitium of the skin, which may have clinical relevance. In addition, volume-regulated anion channels enable cells to discharge osmotic molecules to the interstitium to protect intracellular fluid volume when body water tonicity is low. Balancing the body's potassium (mostly intracellular) with sodium (mostly extracellular) depends on an adequate availability of magnesium. Rapid extracellular fluid osmolality changes can dangerously disturb the intracellular–extracellular fluid equilibrium, so awareness of the major contributors to plasma osmolality is essential. However, evidence from surgical practice suggests that adaptive mechanisms exist to stabilise the intracellular volume in the face of excessive intravenous fluid infusions, and an alternative model of body water response to intravenous infusions is proposed. See also Chap. 3 for the second part on fluid physiology.

Total Body Water

The deuterium nuclear magnetic resonance (^{2}H NMR) is a modern method that has been validated for the determination of total body water in humans. The ^{2}H NMR method has advantages over other techniques based on ^2H_2O dilution. It is fast, accurate, needs only a small dose of ^2H_2O, and can be done using any body fluid [2]. Total body water data from the Fels Longitudinal Study (1999) suggest that the average white American woman has around 30 kg of total body water contributing to her body weight of 65–75 kg, while the

average white American man has around 42–44 kg of total body water contributing to his body weight of 75–93 kg [3]. Bioimpedance spectroscopy (BIS) and multifrequency bioimpedance analysis (MFBIA) are alternative research technologies [4]. Direct measurement of body water has yet to find a role in the clinical diagnosis of dehydration [5].

Most tissues contain 70–80% water, the exceptions being bone and adipose tissue at 10–20%. The major contributors to variation in an individual's total body water to weight are muscle mass (high in water content) and adiposity (fat is low in water content). This explains why females and older individuals typically have a lower percentage of total body water to body weight and may be more prone to disorders of tonicity. As a rule of thumb, the total body water can be estimated as 50% of body weight. In very muscular adults, total body water may be greater than the average. For greater precision, there are a number of anthropomorphic equations for the calculation of total body water. The Watson formula for total body water was derived from and validated on several hundreds of patients. It uses height, weight, age and gender:

- Women Total body water = $-2.097 + 0.1069 \times \text{Height} + 0.2466 \times \text{Weight}$.
- Men Total body water = $2.447 - 0.09156 \times \text{Age} + 0.1074 \times \text{Height} + 0.3362 \times \text{Weight}$.

Clinical Use of Total Body Water Estimation and Modified Body Weights

The rational fluid prescriber is greatly assisted by an estimate of the patient's total body water to make measured decisions about solvent/solute imbalances and the doses of fluid and/or electrolytes needed to correct them. Total body water estimates are also needed to decide the appropriate doses of hydrophilic drugs. For non-obese patients, total body water is proportionate to body weight, but with increasing obesity the utility of body weight as a scalar of total body water and cardiac output diminishes; the excess weight is predominantly fat rather than water and takes little of the cardiac output. Anaesthetic muscle relaxant drugs and antibiotics are hydrophilic and the preferred scalar of dose should logically be total body water. In anaesthetic practice, the recommended dose of hydrophilic drugs such as non-depolarising muscle relaxants is usually scaled to body weight for the non-obese, and to the ideal body weight for obese patients. The Devine formula is widely used:

- Women: Ideal Body Weight (in kilograms) = 45.5 + 2.3 kg/in. over 5 ft.
- Men: Ideal Body Weight (in kilograms) = 50 + 2.3 kg/in. over 5 ft.

Ideal body weight is the best-adjusted weight for following total body water. It is reasonable to presume that the volume of distribution of urea is the total body water and thereby be able to titrate haemofiltration or haemodialysis prescription by ideal body weight to the desired rate of change of urea concentration or to clear other toxins.

An **estimated lean body mass** formula (eLBM) for the normalisation of body fluid volumes was proposed in 1984, the Boer formula [6].

- Women: eLBM = 0.252W + 0.473H − 48.3.
- Men: eLBM = 0.407W + 0.267H − 19.2.

The Peters formula has been proposed for use in anaesthesia and critical care of boys and girls 14 years or younger. The formula first calculates the estimated extracellular fluid volume and then derives the eLBM [7].

- Estimated extracellular fluid volume (eECV) = $0.0215 \times W^{0.6469} \times H^{0.7236}$.
- eLBM = 3.8 × eECV.

Estimated lean body mass is the best scalar of cardiac output in obese patients and is therefore the preferred scalar of the induction dose of hypnotic agents such as di-isopropyl phenol or thiopentone sodium and for the initial dosing of intravenous opiates such as fentanyl and remifentanil. Notice however that these agents are lipophilic and so the measured body weight is the appropriate scalar for setting the steady-state rate of maintenance infusion, even in obese patients.

Tidal volume recommendations for pulmonary ventilation are given in mL/kg **predicted body weight** (PBW), a parameter calculated from height and gender as height most closely predicts normal lung volumes in men and women [8]. In emergency situations when body weight has not been measured or recorded, predicted body weight can rapidly be estimated from height and gender to guide fluid therapy and drug dosing.

Water Absorption

Water is absorbed into the body from the intestinal lumen across the intestinal epithelial membrane and absorption is influenced by luminal osmolality, solute absorption and the anatomical structures of the intestine [9]. Epithelial cells pump intracellular sodium via membrane-bound sodium potassium ATP-ase into the mucosal interstitium, enabling an isosmotic diffusion of water from the lumen into the epithelial cell and thence into the mucosal interstitial fluid. For as long as the epithelium is secreting salt and water to the interstitium, the mucosal capillaries and venules can continue in a steady state of fluid absorption to the plasma. These are diaphragm-fenestrated capillaries, specialised to permit high transendothelial absorption rates. The fenestrations are 10–30 nm wide and formed by the condensation of the luminal and abluminal endothelial cell membranes. They support a diaphragm that functions as a low-resistance filtration/absorption barrier.

Figure 2.2 illustrates the absorption pathway from the intestinal lumen to the plasma. Some interstitial fluid is also removed by lymphatic pumping. Should intestinal water absorption dry up, the epithelial secretion of solvent and small solutes that provides an

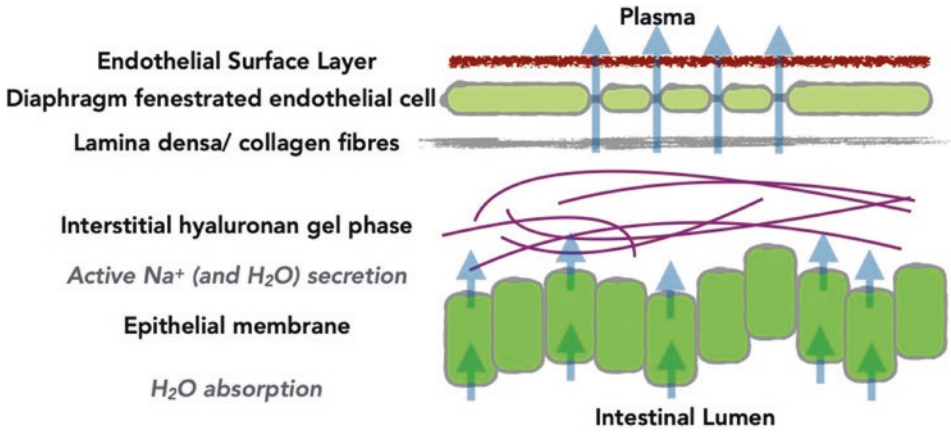

Fig. 2.2 The absorption pathway from intestinal lumen to plasma

exception to the Michel–Weinbaum model of steady-state fluid filtration also ceases, so that microvascular steady-state filtration of plasma solvent to the tissues without reabsorption is restored and gastro-intestinal fluid loss is prevented.

The other tissue microvascular beds in which we find diaphragm-fenestrated capillaries for the absorption of interstitial fluid include exocrine glands, renal peritubular, endocrine glands, peripheral ganglia, nerve epineurium, circumventricular organs, choroid plexus and the ciliary process of the eye.

Plasma Volume

Plasma volume is a critical care concept rather than an anatomic entity. It is often measured as the volume of distribution of an indicator dye such as Evans Blue or radio-labelled albumin, spaces which are substantially larger than the calculated intravascular erythrocyte dilution volume. When reading clinical studies that report plasma volume, it is helpful to bear in mind what has actually been measured or calculated. Broadly, the normal circulating plasma volume is around 2.5–3.0 L at sea level in men and falls by 15–20% with a sojourn to high-altitude living. The circulating plasma contains around 2 L of blood cells (mostly erythrocytes), giving a circulating blood volume at sea level of about 4.5 L [10]. The third contributor to the total intravascular volume is the slower-moving fluid within the endothelial glycocalyx layer, which has been estimated to be as much as 1.5 L in healthy adults or as little as a few hundred milli litres in a variety of cardiovascular disease states [11, 12]. For our approximation we can call it 0.5 L, giving a total intravascular volume of about 5 L at sea level.

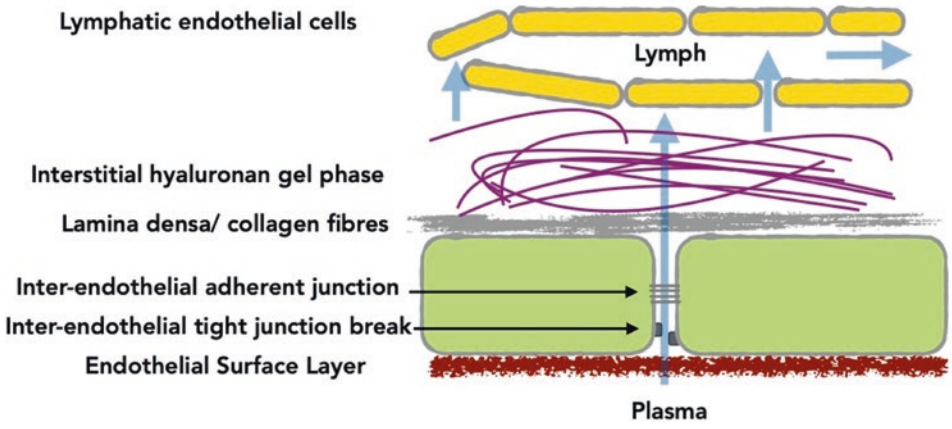

Fig. 2.3 The paracellular (inter endothelial cleft) fluid filtration pathway of continuous capillaries

The circulatory flow rate of blood (Q_t) is supported by the elastic recoil of venules and small veins on the post-capillary blood volume. The venular filling pressure of around 18 mmHg provides a gradient down to the central venous pressure, which is kept close to zero by the healthy heart. The ventricles fill during diastole from the venous excess volume, and the flow to the ventricle is kept nearly constant by the contraction of the atrium. The ventricles eject blood against an afterload that raises the pressure of blood within pulmonary and systemic arteries for distribution to the tissues [13].

Critical care practitioners fear reduced plasma volume with consequent reduced Q_t. They place wide-bore catheters within large veins for 'access' by which to administer vital medicines, and to allow the infusion of 'resuscitation' fluids. In most critical care patients, the intravenous route is the predominant source of fluid input.

The fluid leaves the circulating plasma volume by transendothelial microvascular filtration to the extravascular extracellular fluid of the perfused tissue or organ [14]. Figure 2.3 illustrates the paracellular (inter endothelial cleft) fluid filtration pathway of continuous capillaries.

Traditional clinical teaching has too often ignored the Starling principle and the heterogeneity of the permeability barriers in tissues and organs, which we consider next.

The Starling Principle and Microvessel Heterogeneity

The idea that a single capillary could simultaneously filter fluid from plasma to the interstitial fluid and absorb it back again was always difficult to believe and had never been seen in laboratory experiments. The diagram showing a declining sum of hydrostatic and osmotic pressure differences from the arteriolar entry to the venular exit of a capillary,

with flow reversal as the osmotic pressure difference exceeds the hydrostatic pressure difference, has been called The Diagram to Forget. The capillaries of sinusoidal organs such as liver, spleen and bone marrow receive 25–30% of the left ventricular output via the hepatic artery and portal vein. The splanchnic microvessels are very 'leaky' indeed. They have a discontinuous endothelial surface layer that allows proteins to pass freely in either direction through transendothelial fenestrae and through interendothelial junction gaps so that no osmotic pressure gradient can occur. In such tissues, the unopposed hydrostatic pressure difference ensures that only transendothelial fluid filtration occurs.

In non-sinusoidal tissues, the endothelial surface layer is continuous and largely impermeable to proteins. The paracellular endothelial barrier here presents a layered structure that depends on a continuous activation of signalling pathways regulated by sphingosine-1-phosphate (S1P) and intracellular cAMP. The layers are

- The glycocalyx and its endothelial surface layer
- The junction breaks (or gaps) of the tight junction strand
- The adherens junction

Solvent and small solutes first pass through the resistance of the endothelial surface layer. The greater resistance occurs where fluid is jetted though infrequent slit-like interendothelial tight junction breaks to the interendothelial cleft. The adherens junction provides a third, variable resistance to the paracellular flow of filtrate [15].

Plasma proteins are concentrated on the intravascular aspect of the endothelial surface layer and the filtrate entering the interendothelial cleft is almost protein-free. Soluble interstitial proteins diffuse into the post-glycocalyx solvent filtrate, and the protein concentration in the fluid of the interendothelial clefts will therefore depend on the rates at which the solvent from the plasma filtrate and protein from the interstitium enter the cleft. At high filtration rates, the protein concentration (and colloid osmotic pressure) in the cleft is low, but as the transendothelial filtration rate falls the protein concentration (and colloid osmotic pressure) in the interendothelial cleft rises. The dependence of the colloid osmotic pressure Starling force on the solvent filtration rate within a subglycocalyx 'protected region' of the general interstitial fluid thus ensures that the osmotic forces oppose but, at steady state, do not reverse the flow of the filtrate. This is the Michel–Weinbaum Model, confirmed by experiments published in 2004.

Exceptions to the No-Absorption Rule

We have, however, already mentioned an exception to the steady-state filtration rule in the intestinal mucosal microvasculature. A supply of protein-free solvent to the interstitium by an adjacent secreting epithelium keeps the interstitial colloid osmotic pressure low so that the colloid osmotic pressure difference that drives fluid absorption can be sustained. Similar conditions are found in the peritubular capillaries of the renal cortex and the ascending vasa recta of the renal medulla, which are in a continuous state of absorption of

interstitial fluid because it is continuously secreted by the renal tubular epithelium. In lymph nodes, the interstitial fluid is continuously replenished by the flow of the prenodal lymph with a low protein concentration.

We know that fluids (and water-soluble medicines) injected into the subcutaneous tissue or muscle are rapidly absorbed because the injected volume creates a local exception to the Michel–Weinbaum Model. If Starling's nineteenth century experiments on the absorption of infused fluids from a dog's leg are to be criticised, we can point to his failure to notice that he was creating an artefactual exception, a low-protein lacuna of injected fluid within the interstitium.

Such exceptions aside, in the microcirculation of most tissues the reabsorption of filtered fluid occurs only transiently and only after a substantial disequilibrium of the Starling forces. Tissue fluid balance thus depends critically on the circulation of extracellular fluid through the interstitium and the efficiency of lymphatic pumping in most tissues. Herein, the clinician finds rational approaches to manipulating the distribution of extracellular fluid. Plasma volume can be preserved, and interstitial fluid volume reduced, by reducing the transendothelial solvent filtration rate and by enhancing lymphatic pumping of high protein fluid to the great veins. Biophysical colloid osmotic pressure therapy was once liberally used to support the plasma volume, but advances in physiology and pathophysiology now explain the limitations of that approach.

The Current Understanding of Starling Forces

Capillary hydrostatic pressure P_c is the main driving force of transendothelial solvent filtration (J_v) from plasma to the interstitium. In congestive states elevated P_c increases J_v and oedema ensues if it is not matched by increased lymph flow. Hypovolaemia reduces P_c so that J_v approaches zero; or put another way, fluid cannot leave the hypovolaemic circulation. The clinician can protect the plasma volume by avoiding transient peaks of high P_c by using smaller bolus doses of intravenous fluid. An infusion of the arteriolar constrictor norepinephrine will also protect against increased P_c and J_v. The optimal norepinephrine dose for the arteriolar effect on P_c is less than the dose that raises arterial pressure [11].

Interstitial hydrostatic pressure P_i falls in the first stages of inflammation because of molecular conformational changes in the biomatrix and enhanced lymphatic pumping, and greatly increases J_v before there is any change in capillary permeability.

Plasma colloid osmotic pressure Π_p is by far the largest Starling force at play, but its influence on J_v is modified by the dependence of the subglycocalyx colloid osmotic pressure Π_g on J_v. As J_v falls, Π_g rises and the colloid osmotic pressure difference ($\Pi_p - \Pi_g$) cannot sustain negative J_v (absorption) for more than about 30 min.

The general interstitial fluid colloid osmotic pressure Π_i is now known to have no direct effect on J_v. Interstitial proteins play their part in regulating J_v by diffusing into interendothelial clefts and varying Π_g.

The net rate of solvent filtration from the plasma to interstitium (excluding the glomerular filtration of more than 100 mL/min) is normally just a few mL/min, averaging 0.3–0.4 L/h or 8 L/day. Evidence from the data of Robert Hahn's fluid kinetic studies suggests that under the extreme disequilibrium of rapid isotonic salt solution infusion (50 mL/min), J_v in patients can transiently approach 25–30 mL/min [16].

Starling Forces; Steady State Variations Versus Abrupt Disequilibrium

Clinical researchers who claim to have discredited the Michel–Weinbaum account of the steady-state Starling principle fail to understand that an abrupt change in a Starling force can, if large enough, result in a transient reversal of transendothelial solvent flow. Acute reduction in capillary hydrostatic pressure or acute elevation of plasma colloid osmotic pressure can result in the absorption of interstitial fluid into the plasma. The well-recognised phenomenon is called autotransfusion, and in adults could be as much as 0.5 L. It is, however, soon followed by a return to steady-state filtration. Pulmonary microcirculation is an obvious example for the clinician of steady-state filtration sustained even at low capillary pressures.

Interstitium and Lymphatics

It has long been appreciated that, at a steady state, the quantity and pressure of the extracellular fluid are just as important in determining plasma volume as is the quantity of plasma protein. A decrease in the volume of the circulating red blood cells is normally compensated by an increase in plasma volume, and an increase in the volume of the circulating red cells is compensated by a decrease in plasma volume. The infusion of hyperosmotic albumin solution causes a transient rise in plasma volume, and the removal of plasma protein causes a temporary decrease in plasma volume. The changes in plasma volume produced by varying the amount of circulating plasma protein are not permanent in normal subjects because the body is able to add protein to the bloodstream or remove it. When the blood volume is raised by the addition of protein, the body withdraws protein from the circulation. When the volume is decreased by lowering the quantity of the circulating protein, the body adds protein to the bloodstream. In conditions of increased body water, the plasma volume rises while plasma protein concentration falls [17].

Extracellular fluid circulates. In health the skeletal muscles (female or male) account for 2.2 or 3.5 L, connective tissues 2.4 or 3.0 L, skin 2.0 or 3.0 L, adipose tissue 2.0 or 1.6 L and nervous system 1.4 and 1.5 L. In either sex, there is roughly a litre of fluid in each of the bone, bone marrow and transcellular spaces. The water in the adipose tissue is predominantly extracellular, so the proportion of total body water that is extracellular

tends to be higher in women and in the obese. In morbid obesity, 50–60% of the total body water may be extracellular.

The basement membrane, where it exists, is a specialised part of the extracellular matrix 60–100 nm in thickness, composed of type IV collagen and laminin and closely adherent to the cell membrane. It imposes some resistance to the circulation of extracellular fluid as it leaves the interendothelial cleft and enters the interstitium.

The interstitium is the extracellular matrix within which reside tissue parenchymal cells. It accounts for about one-sixth of the total body volume. The perivascular extracellular matrix forms an organ-specific vascular niche that orchestrates mechano-, growth factor and angiocrine signalling required for tissue homeostasis and organ repair. The composition of the interstitium is controlled by the regulation of synthesis and turnover of each of its individual components, driven by cytokines and growth factors [18]. Around half of the total body albumin is circulating through the interstitium at any one time, and this proportion increases in critical illness as the transendothelial albumin transfer rate from plasma to interstitium increases. This accelerated transfer of albumin is the major cause of post-surgical or post-traumatic hypoalbuminaemia.

Interstitial fluid traverses three 'phases' of the interstitium. The fact of an extravascular circulation of extracellular fluid draws our attention to the way interstitium channels the flow. The major structural elements of the interstitium are collagen fibre bundles, which are visible to light microscopy and can extend for long distances. Probe-based confocal laser endomicroscopy (pCLE) is an in vivo imaging technology that provides real-time histologic assessment of tissue structures in patients. The technology has recently been used to visualise the interstitium of the gastrointestinal tract and urinary bladder submucosae, the dermis, peri-bronchial and peri-arterial soft tissues and fascia. The interstitial space is generally defined by a complex lattice of thick collagen bundles that are intermittently lined on one side by fibroblast-like cells that can be stained with endothelial cell markers. These cell-lined collagen bundles channel the circulation of interstitial fluid.

The Triphasic Interstitium; Collagen Phase

For the purposes of understanding interstitial water disposition, the interstitium has been described as triphasic [19]. The collagen triple helix consists of three intertwined polypeptide chains that entangle water molecules, a property called collagen hydration. Collagen fibre bundles can therefore be considered one of the interstitial aqueous phases. Collagen bundles of several interstitial spaces have been reported to be associated with thin, flat cells (spindle-shaped in cross section) that have scant cytoplasm and an oblong nucleus, and express the transmembrane phosphoglycoprotein CD34. These cells lack the ultrastructural features of endothelial differentiation yet appear to channel the flow of interstitial fluid [20]. Endothelial cell membrane-bound integrins can act upon collagen fibrils in

the adjacent (perivascular) extracellular matrix, exposing glycosaminoglycans (GAGs) to take up water and thereby lower the interstitial pressure.

The Triphasic Interstitium; Hyaluronan Gel Phase

The interstitial gel phase is largely composed of coiled and twisted proteoglycan filaments, barely visible on electron microscopy but holding 99% of the interstitial water in association with glycosaminoglycans, mostly hyaluronan. Hyaluronan restricts the movement of water and forms a diffusion barrier that regulates the transport of substances through intercellular spaces. Hyaluronan takes part in the partitioning of plasma proteins between vascular and extravascular spaces, and creates the excluded volume phenomenon that affects the solubility of macromolecules in the interstitium, changes chemical equilibria, and stabilises the structure of collagen fibres. Interstitial water and solutes of the gel phase occupy the spaces within the proteoglycan/ hyaluronan matrix. The effective radius of these spaces, known as their hydraulic radius, is as small as 3 nm in cartilage and up to 300 nm in the vitreous body of the eye. The hydraulic radius of a matrix determines its resistance to the flow of solvent and solutes through that part of the interstitium. The Wharton's Jelly of the umbilical cord is an open and loosely organised matrix with a hydraulic radius of about 30 nm and is a good example of the gel nature of the interstitium. The interstitial gel restricts water mobility and so stabilises tissue shape. It also prevents interstitial fluid displacement by gravity and slows the spread of organisms such as bacteria. Interstitial hyaluronan washout when lymph flow is raised during systemic inflammation could well contribute to elevated plasma hyaluronan concentrations which are most commonly attributed to disturbance of the endothelial glycocalyx.

Toll-like receptors are found within the extracellular matrix and are believed to have a pivotal role in the early development of systemic inflammatory response and ventilator-induced lung injury. Integrins and their receptors modulate cell locomotion through the extracellular matrix, and can also modulate the interstitial pressure.

The Triphasic Interstitium; Aqueous Phase

Around 1% of interstitial water is normally within a gel-free phase through which water can flow alongside collagen fibre bundles with their associated CD34+ interstitial cells. This space appears microscopically as fluid vesicles and rivulets. The proportion of the gel-free water phase is increased in interstitial oedema, and in the most severe cases up to 50%. Interstitial gel-free solvents and solutes are drawn into collecting lymphatics in order to complete the circulation of interstitial fluid.

Gel Swelling Pressure

GAGs attract water molecules and confer the ability of the interstitial gel phase to swell by taking up water. The gel swelling pressure is defined as the subatmospheric pressure that precisely balances the suction effect of the interstitial GAGs, and is an osmotic pressure largely due to sodium ions attracted by the fixed negative charges—the Gibbs–Donnan effect. Many tissues maintain a subatmospheric interstitial fluid pressure in health because the GAGs are normally under-saturated with water. The lymphatic system maintains this state of under-saturation by pumping fluid away from the gel phase of the interstitium into the aqueous lymph. Reduction of the pumping capacity of the lymphatic system therefore predisposes to fluid retention and oedema.

In collagen-rich tissues such as the skin, the swelling tendency of the interstitial matrix is further counteracted by tissue fibroblasts which tension the collagen fibrils under the regulation of collagen-binding integrins at the cell membrane contact points. The tension of collagen fibres restricts the swelling of GAGs. Collagen-binding integrins only have a limited role in adult connective tissue homeostasis because of the relative paucity of cell-binding sites in the mature fibrillar collagen matrices. Their importance may be greater in connective tissue remodelling, such as wound healing. The skin has been recognised to hold a substantial non-osmotic store of sodium within its interstitium, with a regulatory role in salt and water homeostasis.

Interstitial Starling Forces

Aqueous interstitial fluid can be harvested from nylon wicks implanted subcutaneously, for instance in the arm and leg. In a study of anaesthetised children, the mean plasma colloid osmotic pressure was 26 mmHg while the sampled interstitial fluid colloid osmotic pressure was about 14 mmHg. Albumin is largely excluded from the interstitial gel phase and the collagen phase but is present in the free-flowing interstitial aqueous phase and in the lymph within lymphatic vessels. Water and small solutes (Na^+, Cl^-, and urea) move easily between aqueous and gel phases, and between intracellular fluid and extracellular fluid according to prevailing osmotic, hydrostatic and electrochemical forces. The presence of proteins will affect the viscosity of the flow and accumulation of water in hypoproteinaemic oedematous conditions. As interstitial fluid accumulates and the aqueous phase expands relative to the gel phase, this mechanism becomes increasingly relevant.

Interstitial fluid volume, and so pressure, varies from tissue to tissue and with the rate of fluid exchange. Integrin activation and subsequent conformational changes to collagen allow the GAGs to become hydrated. This brings about an acute reduction in interstitial fluid pressure in inflammatory conditions, increasing the transendothelial pressure difference and thereby increasing J_v by as much as 20-fold independently of other causes of capillary leak.

Water absorption from the gut lumen is associated with the increased mucosal interstitial fluid pressure that promotes water transfer to the plasma by fenestrated mucosal capillaries. Fluid secretion, for instance by endocrine and salivary glands, reduces their interstitial pressure and so increases transendothelial filtration to supply the water needed to continue the secretion. The matrix compressive effect of fibroblasts via collagen-binding integrins has only a limited effect on the regulation of interstitial fluid pressure.

Lymphatic Vascular System

St George's Hospital surgeon William Hunter demonstrated the role of lymphatics in absorbing tissue fluids to the bloodstream in the mid-eighteenth century, yet in clinical teaching today the role of the lymphatic vascular system is often misrepresented by calling it a drainage system. The lymphatic system pumps fluid from tissues and returns it to blood vessels. Lymphatics also transport lymphocytes and dendritic cells to the lymphoid organs. The lymphatic system vasculature consists of thin-walled capillaries and larger vessels that are lined by endothelial cells [21]. There are unique lymphatic markers that differentiate lymph vessels from blood vessels. These include Prox1, a transcription factor required for programming the phenotype of the lymphatic endothelial cell, and LYVE-1, a CD44 homologue. Vascular endothelial growth factor receptor 3 is a receptor for vascular endothelial growth factors (VEGF) C and D, and is not detected in blood vascular endothelial cells. VEGF-C and VEGF-D regulate lymphangiogenesis by activating VEGFR-3, a cell-surface tyrosine kinase receptor, leading to the initiation of a downstream signalling cascade.

The afferent lymphatic vessels are of two types**,** initial and collecting. They differ anatomically (i.e. the presence or absence of surrounding smooth muscle cells and semilunar lymphatic valves), in their expression pattern of adhesion molecules and in their permissiveness to fluid and cell entry. A lymphangion is defined as the functional unit of a lymph vessel that lies between two lymphatic valves.

The afferent lymphatics deliver around 8 L of lymph to lymph nodes per day. The colloid osmotic pressure of lymph is substantially lower than Π_p and its continuous delivery to the lymph node creates a Starling principle exception, allowing the absorption of solvent to the plasma, about 4 L/day, to be sustained. The remaining 4 L/day of lymph proceed to the efferent system.

Efferent lymphatic vessels conduct lymph away from lymph nodes, to further lymph nodes or the lymphatic trunks. They also feature semilunar valves to ensure one-way flow and an investment of smooth muscle to pump the contained fluid. The right and left lumbar trunks and the intestinal trunk constitute the cisterna chyli. The left lymphatic duct, more often called the thoracic duct when seen in the chest, originates on the 12th thoracic vertebra from the confluence of the right and left lumbar trunks, then traverses the diaphragm at the aortic aperture and ascends the superior and posterior mediastinum between the descending thoracic aorta and the azygos vein. The left lymphatic duct averages about

5 mm diameter as it passes behind the left carotid artery and left internal jugular vein at the fifth thoracic vertebral level and drains into the venous angle of the left subclavian and internal jugular veins. There are two valves at the junction of the duct with the left subclavian vein that prevent the flow of venous blood into the duct when central venous pressure exceeds thoracic duct lymph pressure. Efferent lymph from the right thorax, right arm, head and neck is conducted by the smaller right lymphatic duct.

The terminal section of the thoracic duct can be examined at the bedside by 2D ultrasound using high-resolution linear probes (7–12 MHz). Anatomic variations were noted in 27% of subjects in a clinical series of several hundred patients. The normal thoracic duct diameter is about 2.5 mm, independent of the subjects' age. The diameter is substantially increased in subjects with congestive heart failure and liver cirrhosis. Dynamic imaging of the chyle flow and valve function was possible. This technology holds promise for future clinical research [22].

A non-muscular lymphatic endothelial vessel network in the dura mater of the mouse brain was discovered by researchers in Helsinki. The dural lymphatic vessels absorb cerebrospinal fluid from the adjacent subarachnoid space and brain interstitial fluid via the glymphatic system described by Iliff [23]. The traditional view of cerebrospinal fluid (CSF) circulation is that it is produced in the choroid plexus, flows slowly through the subarachnoid space, and is reabsorbed by arachnoid villi or around spinal nerves. In the new paradigm, CSF is a fluid with tightly controlled chemical constituents that flows rapidly around the subarachnoid space and through the brain and spinal cord tissue, fulfilling a role similar to the lymphatic system in other organs. A significant portion of CSF enters the brain via para-vascular spaces (Virchow–Robin spaces) that surround penetrating arteries and arterioles (periarterial spaces). The fluid then leaves the brain via peri-venular spaces. This fluid flow is facilitated at least in part by aquaporins in the end feet of astrocytes, which surround the brain vasculature and form a key component of the blood–brain barrier. Functional lymphatic tissue has been found lining the dural sinuses, which conduct fluid into deep cervical lymph nodes via foramina at the base of the skull, where solvent and small solutes can be absorbed to lymph node venules while efferent lymph flows to the right thoracic duct.

The renal medulla has no lymphatic vessels. Fluid that is absorbed from the collecting ducts into the renal medullary interstitium must therefore be continuously absorbed into the bloodstream by the ascending vasa recta capillaries. It has been demonstrated that labelled albumin is cleared from the medullary interstitium directly into the blood, and it has been calculated that the convective flow of large solutes can account for this efficient clearance.

Interstitial Fluid Dynamics

Intrinsic lymphatic pumping is regulated by four major factors: preload, afterload, spontaneous contraction frequency and contractility. The similarity to the Frank–Starling relationship for the heart is obvious.

- **Preload** is the end-diastolic pressure (or volume) within the valved muscular lymphangion. Increasing the 'filling pressure' over a physiologic range increases the **amplitude of contraction** and so enhances pump output.
- **Afterload**: The lymphatic pump must adapt to elevated outflow pressures resulting from partial outflow obstruction, increased central venous pressure and/or gravitational shifts. Lymphangions in series can propel lymph against higher pressures than individual lymphangions.
- **Contraction frequency** of collecting lymphatics is exquisitely sensitive to pressure, and changes as small as 0.5 cm H_2O can double the contraction frequency.
- **Contractility** is often used in the lymphatic context to describe the enhancement of amplitude or frequency of contraction in response to a pressure increase or agonist activation. The cardiac parallel is the concept of inotropy and inotropic agents.

There are of course extrinsic pump mechanisms operative in vivo. Leg muscles, for example, contribute significantly to the energy expended on pumping lymph to the inguinal, femoral and iliac lymph nodes. Lymph flow in the thoracic duct is supported by the cycle of breathing. The thoracic duct smooth muscle is capable of contracting with sufficient force to propel lymph towards the jugular venous junction at 1–3 mL/min which is just about sufficient to move the normal daily efferent lymph volume of around 4 L.

Lymphatic muscle contractions, like cardiac muscle contractions, can occur spontaneously, but in health, they are subject to neural modulation. Sympathetic adrenergic nerve fibres appear to be the dominant neural innervation of the lymphatic vasculature. α-adrenergic stimulation of contractile lymphatic vessels consistently increases tone, amplitude and frequency, while β-adrenergic receptor activation decreases them. Substance P, commonly associated with afferent nerve endings, augments tone and increases frequency. Muscarinic receptors promote an increase in frequency, but the inhibitory effect of endothelial nitric oxide synthase (eNOS) activation seems to be predominant. Mu receptor agonists such as endorphins and morphine reduce the spontaneous contractility of smooth muscle everywhere. Serotonin (5-HT) can either inhibit or increase spontaneous lymphatic contractions depending on the species and the state of serotonin receptor expression. Other inhibitory factors include vasoactive intestinal peptides and calcitonin gene-related peptides.

Contraction synchrony within a lymphangion generates a systolic pressure pulse that can open the outflow valve and eject lymph. Lymphatic contractions are triggered by an action potential achieved in a pacemaker lymphatic microvascular cell, and the action potential propagates rapidly from cell to cell over the length of the lymphangion. Electrical coupling between the cells is presumably through connexins that form intercellular gap junctions. Application of gap-junction blockers in mesenteric lymphatic vessel segments leads to uncoordinated contractions.

Valve function is critical. Collecting lymphatics contain bicuspid (semilunar) valves whose leaflets extend from a ring-shaped base and insert into the vessel wall. The valve opening is a tapered funnel. A dilated sinus downstream from the valve facilitates valve opening and partially balances the high resistance of the narrow orifice created by the

valve leaflets. Valves are spaced at semi-regular intervals, and the factors that control their spacing are not known.

Barrier function of lymphatic vessels was once disregarded, presuming they were impermeable to fluid and solute. More recent analyses of collecting lymphatic endothelial junction proteins reveal no major differences from those of blood vessels. Collecting lymphatics are not only permeable to solute and fluid, their albumin permeability is comparable to that of post-capillary venules. Like venules, lymphatic permeability is actively regulated because it can be modified by several signalling pathways, including nitric oxide. Lymphatic capillaries are an order of magnitude more permeable than collecting lymphatic microvessels, most likely due to their discontinuous pattern of junctional adhesion proteins, facilitating fluid and solute absorption from the interstitium.

Lymphatic contractile dysfunction is often contingent on inflammatory states such as trauma, sepsis, burns and even major surgery. It is a likely contributor to the accumulation of interstitial fluid or oedema seen in these conditions.

Lymphatics and the Interstitial Storage of Sodium

It has long been taught that body sodium content directly determines the extracellular fluid volume and therefore the effective circulating fluid volume. Long-term blood pressure regulation, it was taught, relies on renal mechanisms to retain or excrete sodium in order to keep the effective circulating fluid volume within very narrow margins of equilibrium. Clinicians therefore use isotonic salt solutions to resuscitate patients with reduced effective circulating fluid volume (hypovolaemia) and are cautioned that excessive sodium administration must cause oedema. Recent investigations in humans confirm animal laboratory evidence that some sodium is in fact stored within the body without commensurate water. This phenomenon was observed with salt solution infusions in surgical patients as long ago as 1986. Indeed, it appears that electrolyte homeostasis in the body cannot be achieved by renal excretion alone, and involves extrarenal regulatory mechanisms such as this. The sodium store is now shown to be an interstitial reservoir that buffers the free extracellular sodium and is regulated by extrarenal, tissue-specific mechanisms for the release and storage of sodium. Immune cells from the mononuclear phagocyte system, including macrophages and dendritic cells, are the local sensors of interstitial electrolyte concentration. The major anatomic site of this sodium regulation is the interstitium of the skin, with its substantial volume of interstitial fluid and lymphatic vasculature forming a vessel network that can be expanded or reduced according to long-term sodium intake. Skin macrophages and lymphatics are now known to act in concert as systemic regulators of body fluid volume and long-term blood pressure. Interstitial electrolyte concentrations are higher than in blood, and macrophages regulate local interstitial electrolyte composition via a tonicity-responsive enhancer-binding protein which induces vascular endothelial growth factor (VEGF-C) production as tonicity rises. Acting on VEGF Receptor 3, VEGF-C stimulates lymphangiogenesis to extend the capillary network and enhance the

capacity for interstitial fluid clearance. At the same time, VEGF-C stimulates VEGF Receptor 2 on blood capillaries promoting endogenous nitric oxide synthesis and increasing local blood flow. Free sodium ions are thus presented via the bloodstream for renal excretion and the extracellular space is protected from major sodium-induced fluid volume fluctuations [24].

A recent study has shown that fluid leaving the skin as lymph is isosmotic to plasma, even after a high sodium intake, but raises the possibility that the skin can differentially control its electrolyte microenvironment by creating local gradients that may be functionally important [25].

To investigate the effects of sodium intake on the endothelial surface layer, 12 healthy male volunteers were randomised to low sodium (less than 50 mmol/day) or high sodium (more than 200 mmol/day) diets for 8 days. There was no measurable effect on arterial pressure, perfused boundary region (endothelial surface layer thickness) or glycosaminoglycan excretion. Body weight increased by around 2.5 kg with high salt intake, suggesting an extracellular volume expansion. Plasma volume measured by the central volume of distribution of radiolabelled albumin was unaffected. Subjects who had followed a low sodium diet were then given 540 mL of 2.4% (hypertonic) sodium chloride as an acute sodium load. This challenge increased the volume of distribution of albumin by 250 mL and increased the transcapillary escape rate of albumin from 7% to 10% per hour. There was no acute effect on arterial pressure or perfused boundary region. The authors' interpretation of their data was that acute intravenous sodium loading was associated with increased microvascular permeability, suggesting functional damage to the endothelial surface layer, but there are other plausible interpretations, including a natriuretic peptide effect. In the same experiment plasma sodium concentration at the end of hypertonic saline infusion was as predicted by standard sodium kinetics, but 4 h later had decreased by 1.8 mmol/L against a predicted fall of less than 1 mmol/L. The authors therefore concluded that healthy individuals are able to osmotically inactivate significant amounts of sodium after hypertonic saline infusion.

Interstitial Fluid and Lymph in Critical Illness

Cope and Litwin (1962) were perhaps the first to demonstrate that fluid absorption to the plasma after acute haemorrhage (reduced capillary pressure) did not fully explain the observed restoration of plasma volume [26]. That volume largely came from a rise in thoracic duct lymph flow, and over the following 24 h lymph flow had returned about twice the amount of protein that had been lost by haemorrhage. They called this phenomenon 'the essentiality of the lymphatic system to the recovery from shock'. Plasma volume refill after blood loss was measured in the 1960s, but is rarely considered in current teaching. Values of 1 mL/min or more imply that the efferent lymph flow is at least doubled during plasma volume refill. Robert Demling in Boston made important

contributions to the pathophysiology of oedema [27]. We see in his work an appreciation of the abrupt disequilibrium of Starling forces moving to a steady state. His laboratory demonstrated that a marked increase in fluid flux after sustained protein depletion is unrelated to colloid osmotic pressure. They drew attention to the possible contribution of decreasing viscosity of the interstitial matrix leading to a more rapid interstitial fluid accumulation. The surgical research team in Denver, Colorado, have developed a hypothesis that mesenteric ischaemia/reperfusion primes polymorphonuclear leucocytes which can then be provoked, for example by endotoxin, to cause distant organ injury by migrating across the endothelium cell and releasing reactive oxygen species [28]. The gut-lymph hypothesis is a variant; the shock-injured gut releases biologically active factors into mesenteric lymph, and these factors activate circulating neutrophils to injure distant endothelial cells.

We now have a dynamic model of an extracellular fluid circulation of solvent, small solutes and albumin in various tissue beds, driven by the lymphatic pump, contributing to the supply and removal of larger, less diffusible molecules to the cells and their intracellular fluid compartment. I summarise this in Fig. 2.4. The relative sizes of the fluid compartments as illustrated here are not to scale with actual volumes.

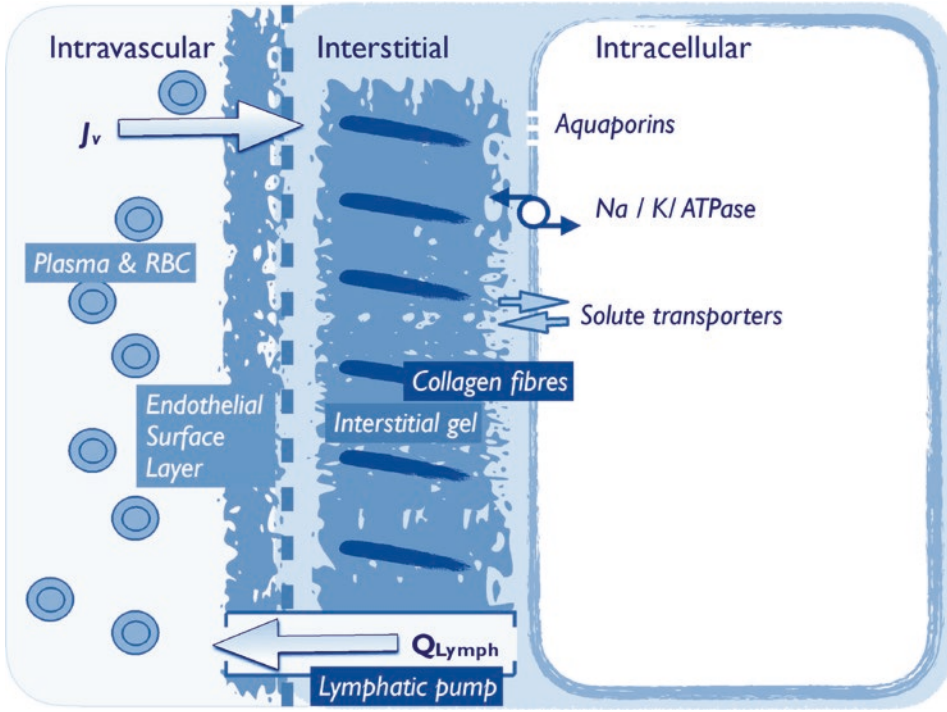

Fig. 2.4 Dynamic model of an extracellular fluid circulation of solvent, small solutes and albumin in various tissue beds

Pulmonary Starling Forces and the Extravascular Lung Water

The pulmonary circulation operates at much lower hydrostatic capillary pressure than the various systemic loops but sustains steady-state solvent filtration to the pulmonary interstitium. As the lungs reside in a sub-atmospheric pressured body space, the interstitial fluid pressure there is lower than that in subcutaneous tissue and fluid from pulmonary capillaries is primarily drawn into the perivascular interstitial space by virtue of the subpleural pressure there. Fluids and solutes are largely excluded from the alveoli by the tight junctions of the alveolar epithelium.

In the progression of pulmonary oedema, fluid accumulates first in the interstitial space around the airways forming 'peribronchial cuffs'. The Staverman reflection co-efficient sigma for albumin in pulmonary capillaries is, on average, around 0.7. There is thus more interstitial protein in the lung than in other tissues with continuous capillaries, and the colloid osmotic pressure difference only weakly opposes steady-state filtration at low capillary pressure. Bronchial artery-supplied capillaries also filter fluid to the pulmonary interstitium and so contribute to the interstitial fluid volume and pulmonary lymph flow.

The volume of pulmonary interstitial fluid is strictly controlled by the lymphatic system. Extravascular lung water can be measured at the bedside by double indicator dilution or by thermodilution alone but includes intracellular fluid. The normal value is about 0.5 L. In severe pulmonary oedema values of more than 1.5 L are recorded, almost all of the excess being extracellular fluid in the interstitium or alveoli.

Cell Fluid and Extracellular Fluid

There are of course cellular elements in the blood which, in females or males, account for 1.0–1.4 L of the intracellular fluid. Skeletal muscle intracellular water varies greatly with muscle mass, but we may nominally expect 11.5 or 18.2 L. The nervous system accounts for 2.5 or 2.8 L. Bone and bone marrow have barely 0.5 L of intracellular fluid, similar to the adipose tissue. Connective tissues and transcellular fluid have very few cells and so very little intracellular water.

Starling Forces Between Extracellular and Intracellular Fluids

Body water distribution between the extracellular and intracellular compartments in each tissue reflects a steady state of hydrostatic pressure and osmosis. At equilibrium, the difference between the intracellular pressure and extracellular pressure is equal and opposite to the osmotic pressure difference across the cell membrane. The magnitude of diffusive water flux due to the osmotic pressure difference of an impermeable solute across an ideal membrane is proportional to the solute's concentration

difference and the membrane's hydraulic conductance (L_p). In reality, the cell membranes are less than ideal barriers, and most solutes are not fully impermeable. A fraction of the partially impermeable solute molecules will therefore be washed through the permeability barrier with solvent flux; this is the convective transport of solutes. In the 1950s Staverman proposed the reflection coefficient sigma (σ) to account for the observed osmotic pressure gradient relative to the ideal osmotic pressure gradient for an impermeable solute. A solute whose sigma approaches zero exerts almost no osmotic pressure (an ineffective osmol), and a solute whose sigma approaches 1 is almost fully effective. Albumin and urea are examples of important solutes whose σ for cell membranes approaches 1 in health, and Staverman's reflection coefficient σ for a solute can be thought of as the fraction of molecules that are reflected by the membrane. When almost all the molecules are reflected σ approaches 1.0. When half of the molecules are reflected, σ is 0.5, and when only one in ten molecules is reflected σ is 0.1.

Maintenance of the Extracellular-Intracellular Solute Balance is Energy-Dependent

Cells need a near-continuous supply of adenosine tri-phosphate (ATP) to extrude permeable Na^+ ions (via membrane channels) which are then balanced by an influx of permeable K^+ ions; sodium and potassium therefore behave like impermeable effective osmoles sequestered in the ECF and ICF. Magnesium is an important co-factor. The sodium-potassium pump was discovered in 1957 by the Danish scientist Jens Christian Skou, a Nobel Prize winner in 1997. For every ATP molecule consumed, three sodium ions leave the cell and two potassium ions enter; there is thus a net export of a positive charge per cycle creating a membrane potential. Chloride (Cl^-) concentrates in the ECF, while fixed anions predominate in the ICF. The fixed intracellular anions include;

- Metabolites such as ATP, phosphocreatine and sulphate
- Nucleotides
- Proteins, which provide most of the intracellular anionic equivalence

Along with potassium they create the Donnan effect osmotic gradient which would draw water into the cell were it not for the **double Donnan effect** of sodium potassium ATP-ase.

Double Donnan Effect

- The intracellular protein concentration (non-diffusible anion) is higher than extracellular, bringing about the first Gibbs–Donnan equilibrium. With unequal distribution of diffusible ions and electric charge, water tends to move into cells.

- Active extrusion of sodium by Na-K pump makes sodium the major extracellular cation, and it has low membrane permeability. This brings about a second Gibbs–Donnan equilibrium that tends to move water out of cells.
- At steady state the two effects balance out and cell volume remains stable, but if sodium potassium ATP-ase is inhibited cells will swell and rupture due to the first Gibbs–Donnan equilibrium. Water is therefore passively distributed between intracellular and extracellular compartments in proportion to the effective Na^+ and K^+ contents to reach effective osmotic equilibrium (tonicity) and establish cell volume.

Potassium and Magnesium Ions

Potassium is the major intracellular cation and 98% of the total body potassium is intracellular. The plasma levels of potassium and magnesium are generally poor indicators of the whole-body content of these electrolytes which are the major intracellular cations, but deficiency does eventually manifest as reduced plasma concentrations. It is a common clinical experience that hypomagnesaemia limits the ability to normalise plasma potassium by giving potassium supplements. Sometimes hypokalaemia only improves after magnesium has been given. The predominant factor seems to be magnesium's part in the working of several weak inward-rectifier potassium channels found in various isoforms along the renal tubular epithelium. The renal outer medullary potassium channel (ROMK) is the prototypic member of this family, and it plays a central role in the regulation of salt and potassium homeostasis [29]. Intracellular magnesium and poly-amines enter the inward-rectifier potassium channel cytoplasmic pore and plug the potassium permeation pathway, giving rise to the phenomenon of 'inward rectification'. In simple terms, intracellular magnesium blocks what would otherwise be the inward flow of potassium and so the recovery of potassium from the lumen of the distal tubule. When intracellular magnesium is depleted, the block is lifted allowing potassium to be conserved.

Cell Volume Regulation and Intracranial Pressure

The principle that cell volume is closely linked to plasma tonicity is particularly important in the nervous system; as plasma tonicity falls, cells swell. An acute onset (usually in <24 h) of hyponatremia causes severe, and sometimes fatal, cerebral oedema. It takes just a 3% fall in plasma osmolality (from 288 to 280 $mosmol^{-1}$) to bring about a 3% increase in brain cell volume, around 40 mL. As the cranial cavity is a rigid box, the same volume (40 mL) of blood and cerebrospinal fluid must be displaced, and this represents 30% of the normal intracranial fluid volume. When intracranial pressure is raised, or intracranial compliance is low, a rapid fall in plasma tonicity can have grave consequences for cerebral perfusion. Hypertonic salt solution boluses (e.g. 3% sodium chloride or 8.4% sodium

bicarbonate) acutely raise plasma tonicity and thus draw water out of brain cells, allowing the intracranial blood volume and perfusion to increase.

With slower tonicity changes, the brain is protected by adaptive steady-state mechanisms, permitting survival at very low serum sodium concentrations. Adaptation to severe hyponatremia is critically dependent on the loss of organic osmolytes from brain cells. These intracellular, osmotically active solutes contribute substantially to the osmolality of cell water and do not adversely affect cell functions when their concentration changes. The volume-regulated anion channels (VRAC) are members of the superfamily of chloride/anion channels. VRAC are activated by cell swelling and restore cell volume by discharging anionic osmoles to the interstitium. VRAC also play a role in cell proliferation, apoptosis, cell migration and the release of various mediators. They could prove to have an important role in central nervous system pathophysiology.

The adaptation that permits survival in patients with severe, chronic (>48 h' duration) hyponatremia also makes the brain vulnerable to injury (osmotic demyelination) if the electrolyte disturbance is corrected too rapidly. The reuptake of organic osmolytes after correction of hyponatremia is slower than the loss of organic osmolytes during the adaptation to hyponatremia. Areas of the brain that remain most depleted of organic osmolytes are the most severely injured by rapid correction. The brain's reuptake of myoinositol, one of the most abundant osmolytes, occurs much more rapidly in a uremic environment, and patients with uraemia are less susceptible to osmotic demyelination. Cerebral demyelination is a rare complication of overly rapid correction of hyponatremia. The principal risk factors for cerebral demyelination are correction of the serum sodium of more than 25 mEq/L in the first 48 h of therapy, correction past the point of 140 mEq/L, chronic liver disease and prior hypoxic/anoxic episode.

Cell Volume Regulation beyond the Brain

Volume-regulated anion channels (VRAC) are not unique to the central nervous system and may prove to have a pivotal role in cell volume regulation in all cell types. Research into the therapeutic potential of hypertonic saline led to the observation that variations in cell volume have quite profound effects on cellular metabolism and gene expression and could, for example, protect against lung injury in a haemorrhagic shock model. Meta-analysis of human studies confirms the expectation of lower-volume resuscitation from sepsis with hypertonic saline, but with no signal of outcome advantage. Hypertonic sodium is also effective as chloride-free sodium lactate. VRAC activity could explain the finding from clinical experience that neither electrolyte-free water nor potassium solution infusions increase the steady-state intracellular fluid volume. Total body water expansion by intravenous fluid infusions of any tonicity appears to be limited to the extracellular fluid volume. Hessels and her colleagues at Groningen have therefore proposed an 'alternative model' of water, sodium and potassium distribution.

Hessels' Alternative Model of Water, Sodium and Potassium Distribution

It has been taught that an infusion of electrolyte-free water will increase the volume of all compartments of the total body water and reduce osmolarity. In a cohort study of post-surgical patients treated in an Intensive Care Unit with conventional intravenous fluids for 4 days, Hessels and colleagues found that there was a strongly positive accumulation of sodium and total fluid, but a negative balance of electrolyte-free water and potassium [30]. In a sub-study comparing the effects of prescribing potassium to a target of 4.0 mmol/L or 4.5 mmol/L they found that all the excess potassium of the second group was renally excreted. They interpreted these observations as showing that excess fluid in clinical practice results in interstitial expansion (extracellular oedema) while the intracellular volume, where potassium is the dominant osmolar cation, is regulated close to its healthy normal. They speculate that the cytosol is able to clear alternative osmolytes when there is a volume increase by electrolyte-free water infusion, and generate alternative osmolytes when hypertonic saline infusion reduces cell volume. Intracellular volume is thereby conserved in the face of changing body water tonicity. Further research including a broader population of critically ill patients would be interesting. Hessels and colleagues have considered the possibility that excess infused sodium is stored non-osmotically in the skin of patients. Their data confirmed 'Missing extracellular sodium' ions, and 'missing extracellular chloride' too [31].

Studies to identify whether the missing ions are held non-osmotically or shifted to the intracellular fluid are warranted.

Water Excretion

There are of course a number of insensible losses, but the major route of water excretion is renal. The glomerular capillaries operate at a high P_c driving a very high filtration rate (the glomerular filtration rate) of solvent and small solutes from the plasma to the renal tubules. The glomerular capillaries feature open (that is, non-diaphragm) fenestrations that have just enough glycocalyx overhanging the edge to retain albumin in the bloodstream. Of the 120 mL/min that are filtered, barely 1 mL/min leaves the collecting ducts to enter the renal pelvis, ureter and urinary bladder for micturition. The filtration capacity of the glomerular capillaries is therefore nearly matched by the absorptive capacity of the peritubular capillaries of the renal cortex and the ascending vasa recta of the renal medulla. The renal tubules and collecting ducts are the secreting epithelia that provide an independent supply of low-protein solvent to the renal interstitium that creates an exception to the no-absorption rule and allows the diaphragm-fenestrated peritubular capillaries and ascending vasa recta to sustain a high absorption rate. Aldosterone adjusts sodium and potassium secretion/absorption in the tubular fluid emerging from the loops of Henle into the distal

convoluted tubules. Arginine vasopressin is the hormone that makes final adjustments to the water conductance of the collecting ducts.

Take Home Messages

- Quantitative consideration of the patient's body water volumes is essential to the prescription of an appropriate rate and total dose of fluid infusion.
- The distribution of extracellular fluid between plasma and the interstitium is most effectively managed by consideration of capillary pressure P_c, or perhaps more specifically of the venular filling pressure.
- While the colloid osmotic pressure of plasma Π_p has historically been considered to be important, we now know that the colloid osmotic pressure difference that opposes filtration varies substantially with the filtration rate J_v and cannot create a sustained (steady state) absorption of interstitial fluid to the plasma.
- While the healthy blood–brain barrier is close to impermeable, all other capillaries are to a greater or lesser extent leaky to solvents and solutes.
- The sinusoidal tissue capillaries take a quarter of the left ventricular output and are totally leaky, so we must dismiss the oft-taught shibboleth that albumin and plasma proteins are confined to the intravascular space.

References

1. Michel CC, Woodcock TE, Curry FE. Understanding and extending the Starling principle. Acta Anaesthesiol Scand. 2020;64(8):1032–7.
2. Khaled MA, Lukaski HC, Watkins CL. Determination of total body water by deuterium NMR. Am J Clin Nutr. 1987;45:1–6.
3. Chumlea WC, Guo SS, Zeller CM, Reo NV, Siervogel RM. Total body water data for white adults 18 to 64 years of age: the Fels Longitudinal Study. Kidney Int. 1999;56:244–52.
4. Ellegård L, Bertz F, Winkvist A, Bosaeus I, Brekke HK. Body composition in overweight and obese women postpartum: bioimpedance methods validated by dual energy X-ray absorptiometry and doubly labeled water. Eur J Clin Nutr. 2016;70:1181–8.
5. Lacey J, Corbett J, Forni L, et al. A multidisciplinary consensus on dehydration: definitions, diagnostic methods and clinical implications. Ann Med. 2019;51:232–51.
6. Boer P. Estimated lean body mass as an index for normalization of body fluid volumes in humans. Am J Physiol. 1984;247:F632–6.
7. Peters AM, Snelling HL, Glass DM, Bird NJ. Estimation of lean body mass in children. Br J Anaesth. 2011;106:719–23.
8. Brower RG, Lanken PN, MacIntyre N, et al. Higher versus lower positive end-expiratory pressures in patients with the acute respiratory distress syndrome. N Engl J Med. 2004;351:327–36.
9. Shi X, Passe DH. Water and solute absorption from carbohydrate-electrolyte solutions in the human proximal small intestine: a review and statistical analysis. Int J Sport Nutr Exerc Metab. 2010;20:427–42.
10. Young AJ, Karl JP, Berryman CE, Montain SJ, Beidleman BA, Pasiakos SM. Variability in human plasma volume responses during high-altitude sojourn. Physiol Rep. 2019;7:e14051.

11. Woodcock TE, Woodcock TM. Revised Starling equation and the glycocalyx model of transvascular fluid exchange: an improved paradigm for prescribing intravenous fluid therapy. Br J Anaesth. 2012;108:384–94.
12. Woodcock TE. Plasma volume, tissue oedema, and the steady-state Starling principle. BJA Educ. 2017;17:74–8.
13. Woodcock T. Fluid physiology: a handbook for anaesthesia and critical care practice. Cambridge Scholars Publishing; 2019.
14. Levick JR, Michel CC. Microvascular fluid exchange and the revised Starling principle. Cardiovasc Res. 2010;87:198–210.
15. Curry FR, Adamson RH. Tonic regulation of vascular permeability. Acta Physiol (Oxf). 2013;207:628–49.
16. Hahn RG. Volume kinetics for infusion fluids. Anesthesiology. 2010;113:470–81.
17. Warren JV, Merrill AJ, Stead EA. The role of the extracellular fluid in the maintenance of a normal plasma volume. J Clin Invest. 1943;22:635–41.
18. Wiig H, Swartz MA. Interstitial fluid and lymph formation and transport: physiological regulation and roles in inflammation and cancer. Physiol Rev. 2012;92:1005–60.
19. Bhave G, Neilson EG. Body fluid dynamics: back to the future. J Am Soc Nephrol. 2011;22:2166–81.
20. Benias PC, Wells RG, Sackey-Aboagye B, et al. Structure and distribution of an unrecognized interstitium in human tissues. Sci Rep. 2018;8:4947.
21. Scallan JP, Zawieja SD, Castorena-Gonzalez JA, Davis MJ. Lymphatic pumping: mechanics, mechanisms and malfunction. J Physiol. 2016;594:5749–68.
22. Seeger M, Bewig B, Günther R, et al. Terminal part of thoracic duct: high-resolution US imaging. Radiology. 2009;252(3):897–904.
23. Iliff JJ, Goldman SA, Nedergaard M. Implications of the discovery of brain lymphatic pathways. Lancet Neurol. 2015;14:977–9.
24. Wiig H, Luft FC, Titze JM. The interstitium conducts extrarenal storage of sodium and represents a third compartment essential for extracellular volume and blood pressure homeostasis. Acta Physiol (Oxf). 2018;222
25. Nikpey E, Karlsen TV, Rakova N, Titze JM, Tenstad O, Wiig H. High-salt diet causes osmotic gradients and hyperosmolality in skin without affecting interstitial fluid and lymph. Hypertension. 2017;69:660–8.
26. Cope O, Litwin SB. Contribution of the lymphatic system to the replenishment of the plasma volume following a hemorrhage. Ann Surg. 1962;156:655–67.
27. Demling RH, Kramer GC, Gunther R, Nerlich M. Effect of nonprotein colloid on postburn edema formation in soft tissues and lung. Surgery. 1984;95:593–602.
28. Dzieciatkowska M, D'Alessandro A, Moore EE, et al. Lymph is not a plasma ultrafiltrate: a proteomic analysis of injured patients. Shock. 2014;42:485–98.
29. Hadchouel J, Ellison DH, Gamba G. Regulation of renal electrolyte transport by WNK and SPAK-OSR1 kinases. Annu Rev Physiol. 2016;78:367–89.
30. Hessels L, Oude Lansink A, Renes MH, et al. Postoperative fluid retention after heart surgery is accompanied by a strongly positive sodium balance and a negative potassium balance. Physiol Rep. 2016;4
31. Hessels L, Oude Lansink-Hartgring A, Zeillemaker-Hoekstra M, Nijsten MW. Estimation of sodium and chloride storage in critically ill patients: a balance study. Ann Intensive Care. 2018;8:97.